

## I.) AMENDMENTS

### IN THE CLAIMS:

1. (currently amended) A method of preparing a reconstructed non-human mammalian oocyte by transferring a cell or nucleus from germinal or somatic cells into an enucleated host mammalian oocyte, which comprises the steps of:

- a) activating ~~said~~ mammalian host oocyte;
- b) enucleating said activated host oocyte when said activated oocyte is undergoing the expulsion of a second polarbody or when said activated oocyte has recently expelled second polarbody(Tel-II); and
- c) transferring a nucleus from mammalian germinal or somatic cells into said enucleated host oocyte of step b) to obtain a reconstructed mammalian oocyte.

2. (original) The method according to claim 1, wherein said transferred cell or nucleus is at nuclear stage G0, G1, S, G2, or M.

3. (original) The method of claim 1, wherein said germinal or somatic cells of step c) are cultured prior to nucleus transfer.

4. (original) The method of claim 1, wherein said oocyte of step a) is a secondary oocyte (M-II) and said activation is performed by artificial means selected from the group consisting of physical means and chemical means.

5. (original) The method of claim 4, wherein said chemical means is ethanol or ionomycin.

6. (original) The method of claim 4, wherein said physical means is selected from the group consisting of electrical means, thermal means, and irradiation technology.

7. (original) The method of claim 1, wherein step b) is performed after oocytes are cultured for a period of time sufficient to allow for at least partial extrusion of a second polarbody.

8. (currently amended) The method of claim 1, wherein step b) is performed with oocytes in a medium with ~~cytoskeletal~~ cytoskeletal inhibitors.

9. (original) The method of claim 7, wherein step b) is effected by microsurgically removing said second polarbody with a portion of the cytoplasm containing chromosomes surrounding said at least partially extruded second polarbody.

10. (currently amended) A method of reconstituting a non-human mammalian embryo, which comprises the steps of:

- a) activating a mammalian oocyte by artificial or natural means;
- b) enucleating said activated oocyte when said activated oocyte is undergoing the expulsion of a second polarbody or when said activated oocyte has recently expelled said second polarbody(Tel-II);
- c) culturing mammalian germinal or somatic cell prior to nucleus transfer;
- d) transferring a nucleus from said cell of step c) in said enucleated oocyte to obtain a reconstructed mammalian oocyte with a diploid chromosomal content; and
- d) culturing *in vitro* said reconstructed oocyte and/or transferring said reconstructed oocyte into a reproductive tract of a suitable surrogate mother to enable development into a non-human mammalian embryo.

11. (original) The method according to claim 10, wherein said transferred cell or nucleus is at nuclear stage G0 G1, S, G2, or M.

12. (original) The method of claim 10, wherein said oocyte of step a) is a secondary oocyte

(M-II) and said artificial means is physical or chemical means.

13. (original) The method of claim 12, wherein said chemical means is ethanol or ionomycin.

14. (original) The method of claim 12, wherein said physical means is selected from the group consisting of electrical means, thermal means, and irradiation technology.

15. (original) The method of claim 13, wherein step b) is performed after oocytes are cultured for a period of time sufficient to allow for at least partial extrusion of a second polarbody.

16. (currently amended) The method of claim 15, wherein step b) is performed with oocytes in a medium with ~~cytoskeletal~~ cytoskeletal inhibitors.

17. (original) The method of claim 15, wherein step b) is effected by microsurgically removing said second polarbody with a portion of the cytoplasm containing chromosomes surrounding said at least partially extruded second polarbody.

18. (original) The method of claim 17, wherein step c) is effected by introducing a single cell containing a diploid nucleus into said enucleated oocyte by cell fusion or by microinjection.

19. (cancelled)

20. (currently amended) A method for production of a transgenic nonhuman mammalian embryo, which comprises the steps of:

a) activating a mammalian oocyte by artificial or natural means;

b) enucleating said activated oocyte when said activated oocyte is undergoing the expulsion of a second polarbody or when said activated oocyte has recently expelled a second polarbody(Tel-II);

- c) culturing a mammalian germinal or somatic cell prior to nucleus transfer;
- d) transferring a transgenic nucleus from said cell of step c) transfected with a desired DNA construct in said enucleated oocyte to obtain a reconstructed oocyte with a diploid chromosomal content; and
- e) culturing *in vitro* said reconstructed oocyte and/or transferring said reconstructed oocyte into a reproductive tract of a suitable surrogate mother to enable development into a non-human mammalian embryo.

21. (original) The method according to claim 20, wherein said transferred cell or nucleus is at nuclear stage G0, G1, S, G2, or M.

22. (original) The method according to claim 20, which further comprises developing said non-human mammalian embryo into a fetus.

23. (original) The method according to claim 22, which further comprises developing said fetus into an offspring.

24. (currently amended) The method of claim 20, wherein said non-human mammalian embryo develops into a non-human mammal.

25. (currently amended) A non-human transgenic embryo obtained according to the method which comprises the steps of:

- a) activating a mammalian oocyte by artificial or natural means;
- b) enucleating said activated oocyte when said activated oocyte is undergoing the expulsion of a second polarbody or when said activated oocyte has recently expelled a second polarbody(Tel-II);
- c) culturing a mammalian germinal or somatic cell prior to nucleus transfer;
- d) transferring a transgenic nucleus from said cell of step c) transfected with a desired DNA

construct in said enucleated oocyte to obtain a reconstructed oocyte with a diploid chromosomal content; and

e) culturing *in vitro* said reconstructed oocyte and/or transferring said reconstructed oocyte into a reproductive tract of a suitable surrogate mother to enable development into a non-human mammalian embryo.

26. (currently amended) A non-human mammalian transgenic fetus obtained according to the method which comprises the steps of:

a) activating a mammalian oocyte by artificial or natural means;

b) enucleating said activated oocyte when said activated oocyte is undergoing the expulsion of a second polarbody or when said activated oocyte has recently expelled a second polarbody(Tel-II);

c) culturing a mammalian germinal or somatic cell prior to nucleus transfer;

d) transferring a transgenic nucleus from said cell of step c) transfected with a desired DNA construct in said enucleated oocyte to obtain a reconstructed oocyte with a diploid chromosomal content; and

e) culturing *in vitro* said reconstructed oocyte and/or transferring said reconstructed oocyte into a reproductive tract of a suitable surrogate mother to enable development into a non-human ~~embryo~~ mammalian fetus.

27. (currently amended) A non-human transgenic offspring obtained according to the method which comprises the steps of:

a) activating a mammalian oocyte by artificial or natural means;

b) enucleating said activated oocyte when said activated oocyte is undergoing the expulsion of a second polarbody or when said activated oocyte has recently expelled a second polarbody(Tel-II);

- c) culturing a mammalian germinal or somatic cell prior to nucleus transfer;
- d) transferring a transgenic nucleus from said cell of step c) transfected with a desired DNA construct in said enucleated oocyte to obtain a reconstructed oocyte with a diploid chromosomal content; and
- e) culturing *in vitro* said reconstructed oocyte and/or transferring said reconstructed oocyte into a reproductive tract of a suitable surrogate mother to enable development into a non-human ~~embryo~~ mammalian offspring.

28. (currently amended) A method of cloning a non-human mammal ~~animal~~ by cell or nuclear transfer which comprises the steps of :

- a) activating a mammalian oocyte by artificial means;
- b) enucleating said activated oocyte when said activated oocyte is undergoing the expulsion of a second polarbody or when said activated oocyte has recently expelled second polarbody(Tel-II);
- c) transferring a diploid nucleus from said cell of step c) in said enucleated oocyte to obtain a reconstructed oocyte with a diploid chromosomal content; and
- d) culturing *in vitro* said reconstructed oocyte and/or transferring said reconstructed oocyte into a reproductive tract of a suitable surrogate mother to enable development into a ~~non-human~~ mammalian embryo.

29. (original) The method according to claim 28, wherein said transferred cell or nucleus is at nuclear stage G0, G1, S, G2, or M.

30. (original) The method of claim 28, wherein said oocyte of step a) is a secondary oocyte (M-II) and said artificial means is physical or chemical means.

31. (original) The method of claim 30, wherein said chemical means is ethanol or ionomycin.

32. (original) The method of claim 30, wherein said physical means is selected from the group consisting of electrical means, thermal means, and irradiation technology.

33. (original) The method of claim 28, wherein step b) is performed after oocytes are cultured for a period of time sufficient to allow for at least partial extrusion of a second polarbody.

34. (currently amended) The method of claim 30, wherein step b) is performed with oocytes in a medium with ~~cytoskeletal~~ cytoskeletal inhibitors.

35. (original) The method of claim 31, wherein step b) is effected by microsurgically removing said second polarbody with a portion of the cytoplasm containing chromosomes surrounding said at least partially extruded second polarbody.

36. (original) The method of claim 32, wherein step c) is effected by introducing a single cell containing a diploid nucleus into said enucleated oocyte by cell fusion or by microinjection.

37. (original) The method of claim 28, wherein said nucleus or cell of step c) is transgenic or non-transgenic.

38. (currently amended) The method of claim 28, wherein said ~~non-human~~ mammalian embryo develops into a ~~non-human animal~~ an offspring.

## **II. REMARKS**

Claims 1-38 are pending in the instant application, and all of the pending claims are rejected.